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SEQUENCE LISTING

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<120> MUTANT KANAMYCIN NUCLEOTIDYLTRANSFERASE AND A METHOD
OF SCREENING THERMOPHILIC BACTERIA USING THE SAME

<130> 04853.0048-0.0000

<140> 09/697,186
<141> 2000-10-27

<150> JP 309616/1999
<151> 1999-10-29

<160> 20

<170> PatentIn Ver. 2.1

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<212> PRT
<213> Artificial Sequence

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<223> Description of Artificial Sequence: Mutant enzyme
obtained by introduction of point mutation into
wild type KNT gene of *Staphylococcus aureus* and
its expression

<400> 1

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val
1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Glu Ala Glu Phe
50 55 60

Ser His Glu Trp Thr Thr Gly Glu Trp Lys Val Glu Val Asn Phe Tyr
65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Gln Val Glu Ser Asp Trp
85 90 95

Pro Leu Thr His Gly Gln Phe Phe Ser Ile Leu Pro Ile Tyr Asp Ser
100 105 110

Gly Gly Tyr Leu Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala
 115 120 125
 Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe
 130 135 140
 Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr
 145 150 155 160
 Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu
 165 170 175
 Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu
 180 185 190
 Thr Glu Ala Val Lys Gln Ser Asp Leu Pro Ser Gly Tyr Asp His Leu
 195 200 205
 Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu
 210 215 220
 Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg
 225 230 235 240
 His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe
 245 250

<210> 2
 <211> 253
 <212> PRT
 <213> Artificial Sequence

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 <223> Description of Artificial Sequence: Mutant enzyme
 obtained by introduction of point mutation into
 wild type KNT gene of *Staphylococcus aureus* and
 its expression

<400> 2
 Met Lys Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val
 1 5 10 15
 His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
 20 25 30
 Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
 35 40 45
 Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe
 50 55 60
 Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr
 65 70 75 80
 Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Ser Asp Trp
 85 90 95

Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro
 100 105 110
 Gly Gly Tyr Phe Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala
 115 120 125
 Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe
 130 135 140
 Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr
 145 150 155 160
 Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu
 165 170 175
 Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu
 180 185 190
 Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Ser Gly Tyr Asp His Leu
 195 200 205
 Cys Gln Leu Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu
 210 215 220
 Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg
 225 230 235 240
 His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe
 245 250

<210> 3
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 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Mutant enzyme
 obtained by introduction of point mutation into
 wild type KNT gene of *Staphylococcus aureus* and
 its expression

<400> 3
 Met Lys Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val
 1 5 10 15
 His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
 20 25 30
 Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
 35 40 45
 Ser Asp Ile Glu Met Met Cys Val Leu Ser Thr Glu Gly Val Glu Phe
 50 55 60
 Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr
 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Pro Asp Trp
 85 90 95

 Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro
 100 105 110

 Gly Gly Tyr Phe Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala
 115 120 125

 Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe
 130 135 140

 Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr
 145 150 155 160

 Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu
 165 170 175

 Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu
 180 185 190

 Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Pro Gly Tyr Val Gln Leu
 195 200 205

 Cys Gln Leu Val Met Ser Gly Gln Leu Ser Asp Pro Glu Lys Leu Leu
 210 215 220

 Glu Ser Leu Glu Asn Phe Trp Asn Gly Val Gln Glu Trp Ala Glu Arg
 225 230 235 240

 His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe
 245 250

<210> 4
 <211> 35
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> Description of Artificial Sequence: Primer for
 PCR amplification

<400> 4
 gactgtacgg gtaccgcgtt acggcgata tggta 35

<210> 5
 <211> 35
 <212> DNA
 <213> Artificial Sequence

 <220>
 <223> Description of Artificial Sequence: Primer for
 PCR amplification

<400> 5
 gactgtacgc tgcaacatgtat taaca 35

<210> 6
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<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer for PCR amplification

<400> 6
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<210> 7
<211> 35
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer for subcloning of WT

<400> 7
gactgtacgc atatgaatgg accaataata atgac 35

<210> 8
<211> 35
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer for subcloning of KT3-11 and HTK

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<210> 9
<211> 35
<212> DNA
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Primer for subcloning

<400> 9
gactgtacgc tcgagcgtaa ccaacatgat taaca 35

<210> 10
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<212> DNA
<213> Staphylococcus aureus

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| gaa tcg cta gag aat ttc tgg aat ggg att cag gag tgg aca gaa cga Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg 225 230 235 240 | 720 |
| cac gga tat ata gtg gat gtg tca aaa cgc ata cca ttt His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe 245 250 | 759 |
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| <213> Staphylococcus aureus | |
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| His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys 20 25 30 | |
| Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr 35 40 45 | |
| Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Glu Ala Glu Phe 50 55 60 | |
| Ser His Glu Trp Thr Thr Gly Glu Trp Lys Val Glu Val Asn Phe Asp 65 70 75 80 | |
| Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Gln Val Glu Ser Asp Trp 85 90 95 | |
| Pro Leu Thr His Gly Gln Phe Phe Ser Ile Leu Pro Ile Tyr Asp Ser 100 105 110 | |
| Gly Gly Tyr Leu Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala 115 120 125 | |
| Gln Thr Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe 130 135 140 | |
| Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr 145 150 155 160 | |
| Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu 165 170 175 | |
| Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu 180 185 190 | |
| Thr Glu Ala Val Lys Gln Ser Asp Leu Pro Ser Gly Tyr Asp His Leu 195 200 205 | |

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu
 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg
 225 230 235 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe
 245 250

<210> 12
<211> 253
<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of *Staphylococcus aureus* and its expression

<400> 12
Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val
 1 5 10 15

Tyr Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
 20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Glu Ala Glu Phe
 50 55 60

Ser His Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr
 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Ser Asp Trp
 85 90 95

Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro
 100 105 110

Gly Gly Tyr Phe Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala
 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe
 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr
 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu
 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu
 180 185 190

Thr Glu Ala Leu Lys Gln Ser Asp Leu Pro Ser Gly Tyr Asp His Leu
 195 200 205
 Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu
 210 215 220
 Glu Ser Leu Glu Asn Phe Trp Asn Gly Val Gln Glu Trp Ala Glu Arg
 225 230 235 240
 His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe
 245 250

<210> 13
 <211> 253
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Mutant enzyme
 obtained by introduction of point mutation into
 wild type KNT gene of *Staphylococcus aureus* and
 its expression

<400> 13
 Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val
 1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asn Lys Tyr Gly Asp Asp Val Lys
 20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
 35 40 45

Ser Asp Ile Glu Met Met Cys Val Leu Ser Thr Glu Gly Val Glu Phe
 50 55 60

Ser His Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr
 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Gln Val Glu Pro Asp Trp
 85 90 95

Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Ser
 100 105 110

Gly Gly Tyr Leu Gly Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala
 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe
 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr
 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu
 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Leu Val Leu
 180 185 190
 Thr Glu Ala Val Lys Gln Ser Asp Leu Pro Ser Gly Tyr Asp His Leu
 195 200 205
 Cys Gln Leu Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu
 210 215 220
 Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg
 225 230 235 240
 His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe
 245 250

<210> 14

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme
 obtained by introduction of point mutation into
 wild type KNT gene of Staphylococcus aureus and
 its expression

<400> 14

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val
 1 5 10 15His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
 20 25 30Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
 35 40 45Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe
 50 55 60Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr
 65 70 75 80Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Pro Asp Trp
 85 90 95Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Ser
 100 105 110Gly Gly Tyr Leu Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala
 115 120 125Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe
 130 135 140Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr
 145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu
 165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu
 180 185 190

Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Ser Gly Tyr Asp His Leu
 195 200 205

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu
 210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Val Gln Glu Trp Thr Glu Arg
 225 230 235 240

His Gly Tyr Ile Val Asn Val Ser Lys Arg Ile Pro Phe
 245 250

<210> 15
<211> 253
<212> PRT
<213> Artificial Sequence

<220>
<223> Description of Artificial Sequence: Mutant enzyme
obtained by introduction of point mutation into
wild type KNT gene of *Staphylococcus aureus* and
its expression

<400> 15
Met Ser Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val
 1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
 20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe
 50 55 60

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr
 65 70 75 80

Ser Glu Glu Ile Leu Asp Tyr Ala Ser Arg Val Glu Ser Asp Trp
 85 90 95

Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro
 100 105 110

Gly Gly Tyr Phe Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala
 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe
 130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr
 145 150 155 160
 Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu
 165 170 175
 Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Thr Val Leu
 180 185 190
 Thr Glu Ala Val Lys Leu Ser Asp Leu Pro Ser Gly Tyr Asp His Leu
 195 200 205
 Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu
 210 215 220
 Glu Ser Leu Glu Asn Phe Trp Asn Gly Val Gln Glu Trp Thr Glu Arg
 225 230 235 240
 His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe
 245 250

<210> 16
 <211> 253
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Mutant enzyme
 obtained by introduction of point mutation into
 wild type KNT gene of *Staphylococcus aureus* and
 its expression

<400> 16
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 1 5 10 15
 His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
 20 25 30
 Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
 35 40 45
 Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Glu Thr Glu Phe
 50 55 60
 Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr
 65 70 75 80
 Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Pro Asp Trp
 85 90 95
 Pro Leu Thr His Gly Lys Phe Phe Ser Ile Leu Pro Ile Tyr Asp Thr
 100 105 110
 Gly Gly Tyr Leu Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala
 115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe
 130 135 140
 Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Leu Thr
 145 150 155 160
 Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu
 165 170 175
 Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Gly Ala Ser Val Leu
 180 185 190
 Thr Glu Ala Val Arg Gln Pro Asp Leu Pro Pro Gly Tyr Asp His Leu
 195 200 205
 Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu
 210 215 220
 Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Ala Glu Arg
 225 230 235 240
 His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe
 245 250

<210> 17

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme
 obtained by introduction of point mutation into
 wild type KNT gene of *Staphylococcus aureus* and
 its expression

<400> 17

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val
 1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
 20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
 35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe
 50 55 60

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr
 65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Ser Asp Trp
 85 90 95

Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro
 100 105 110

Gly Gly Tyr Phe Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala
 115 120 125
 Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe
 130 135 140
 Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr
 145 150 155 160
 Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu
 165 170 175
 Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu
 180 185 190
 Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Ser Gly Tyr Val Gln Leu
 195 200 205
 Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu
 210 215 220
 Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg
 225 230 235 240
 His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe
 245 250

<210> 18

<211> 253

<212> PRT

<213> Artificial Sequence

<220>

<223> Description of Artificial Sequence: Mutant enzyme
obtained by introduction of point mutation into
wild type KNT gene of *Staphylococcus aureus* and
its expression

<400> 18

Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val
1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
35 40 45

Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe
50 55 60

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr
65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Ser Asp Trp
85 90 95

Pro Leu Thr His Gly Arg Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro
 100 105 110
 Gly Gly Tyr Phe Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala
 115 120 125
 Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe
 130 135 140
 Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr
 145 150 155 160
 Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu
 165 170 175
 Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu
 180 185 190
 Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Ser Gly Tyr Val Gln Leu
 195 200 205
 Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu
 210 215 220
 Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg
 225 230 235 240
 His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe
 245 250

<210> 19
 <211> 253
 <212> PRT
 <213> Artificial Sequence

<220>
 <223> Description of Artificial Sequence: Mutant enzyme
 obtained by introduction of point mutation into
 wild type KNT gene of *Staphylococcus aureus* and
 its expression

<400> 19
 Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val
 1 5 10 15
 His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
 20 25 30
 Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
 35 40 45
 Ser Asp Ile Glu Met Met Cys Val Met Ser Thr Glu Gly Ala Glu Phe
 50 55 60
 Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr
 65 70 75 80

| | | | | | | | | | | | | | | | |
|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|-----|
| Ser | Glu | Glu | Ile | Leu | Leu | Asp | Tyr | Ala | Ser | Arg | Val | Glu | Pro | Asp | Trp |
| | | | | 85 | | | | | | 90 | | | | | 95 |
| Pro | Leu | Thr | His | Gly | Lys | Phe | Phe | Ser | Ile | Leu | Pro | Ile | Tyr | Asp | Pro |
| | | | | | 100 | | | | 105 | | | | 110 | | |
| Gly | Gly | Tyr | Leu | Glu | Lys | Val | Tyr | Gln | Thr | Ala | Lys | Ser | Val | Glu | Ala |
| | | | | | 115 | | | 120 | | | | 125 | | | |
| Gln | Lys | Phe | His | Asp | Ala | Ile | Cys | Ala | Leu | Ile | Val | Glu | Glu | Leu | Phe |
| | | | | | 130 | | | 135 | | | | 140 | | | |
| Glu | Tyr | Ala | Gly | Lys | Trp | Arg | Asn | Ile | Arg | Val | Gln | Gly | Pro | Thr | Thr |
| | | | | | 145 | | | 150 | | | 155 | | | | 160 |
| Phe | Leu | Pro | Ser | Leu | Thr | Val | Gln | Val | Ala | Met | Ala | Gly | Ala | Met | Leu |
| | | | | | 165 | | | | 170 | | | | 175 | | |
| Ile | Gly | Leu | His | His | Arg | Ile | Cys | Tyr | Thr | Thr | Ser | Ala | Ser | Val | Leu |
| | | | | | 180 | | | 185 | | | | 190 | | | |
| Thr | Glu | Ala | Val | Lys | Gln | Pro | Asp | Leu | Pro | Ser | Gly | Tyr | Val | Gln | Leu |
| | | | | | 195 | | | 200 | | | | 205 | | | |
| Cys | Gln | Phe | Val | Met | Ser | Gly | Gln | Leu | Ser | Asp | Pro | Glu | Lys | Leu | Leu |
| | | | | | 210 | | | 215 | | | | 220 | | | |
| Glu | Ser | Leu | Glu | Asn | Phe | Trp | Asn | Gly | Ile | Gln | Glu | Trp | Thr | Glu | Arg |
| | | | | | 225 | | | 230 | | | 235 | | | | 240 |
| His | Gly | Tyr | Ile | Val | Asp | Val | Ser | Lys | Arg | Ile | Pro | Phe | | | |
| | | | | | 245 | | | | 250 | | | | | | |

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<210> 20
<211> 253
<212> PRT
<213> Artificial Sequence
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<220>
<223> Description of Artificial Sequence: Mutant enzyme obtained by introduction of point mutation into wild type KNT gene of *Staphylococcus aureus* and its expression

<400> 20
Met Asn Gly Pro Ile Ile Met Thr Arg Glu Glu Arg Met Lys Ile Val
1 5 10 15

His Glu Ile Lys Glu Arg Ile Leu Asp Lys Tyr Gly Asp Asp Val Lys
20 25 30

Ala Ile Gly Val Tyr Gly Ser Leu Gly Arg Gln Thr Asp Gly Pro Tyr
35 40 45

Ser Asp Ile Glu Met Met Cys Val Leu Ser Thr Glu Glu Ala Glu Phe

Ser Tyr Glu Trp Thr Thr Gly Glu Trp Lys Ala Glu Val Asn Phe Tyr
65 70 75 80

Ser Glu Glu Ile Leu Leu Asp Tyr Ala Ser Arg Val Glu Pro Asp Trp
85 90 95

Pro Leu Thr His Gly Lys Phe Phe Ser Ile Leu Pro Ile Tyr Asp Pro
100 105 110

Gly Gly Tyr Leu Glu Lys Val Tyr Gln Thr Ala Lys Ser Val Glu Ala
115 120 125

Gln Lys Phe His Asp Ala Ile Cys Ala Leu Ile Val Glu Glu Leu Phe
130 135 140

Glu Tyr Ala Gly Lys Trp Arg Asn Ile Arg Val Gln Gly Pro Thr Thr
145 150 155 160

Phe Leu Pro Ser Leu Thr Val Gln Val Ala Met Ala Gly Ala Met Leu
165 170 175

Ile Gly Leu His His Arg Ile Cys Tyr Thr Thr Ser Ala Ser Val Leu
180 185 190

Thr Glu Ala Val Lys Gln Pro Asp Leu Pro Ser Gly Tyr Val Gln Leu
195 200 205

Cys Gln Phe Val Met Ser Gly Gln Leu Ser Asp Ser Glu Lys Leu Leu
210 215 220

Glu Ser Leu Glu Asn Phe Trp Asn Gly Ile Gln Glu Trp Thr Glu Arg
225 230 235 240

His Gly Tyr Ile Val Asp Val Ser Lys Arg Ile Pro Phe
245 250